and participates in a salt-bridge between the switch I and switch II nucleotide binding regions. We observed very minimal actin-activated ATPase activity for the mutant compared to WT (2-fold compared to 100-fold activation, respectively), while the basal ATPase was unchanged. R243H bound to actin in an ATP-dependent manner based on actin-cosedimentation assays. We also observed no actin-sliding by R243H in the in vitro motility assay. In the mixed motility assay, low ratios ($\leq$50%) of R243H did not change or only slightly decreased sliding velocities. At higher ratios of R243H ($\geq$50%) the velocity decreased dramatically. We speculate that at low ratios of R243H/WT the mutant myosin may cooperatively activate the actin thin filaments to allow more opportunity for WT myosin heads to bind actin without slowing shortening velocity, which would correlate with HCM. At higher ratios of R243/WT, the inhibitory action of R243H may dominate and slow muscle shortening, which would correlate with DCM.

1301-Pos

Molecular Dynamics Studies of Single Point Mutations in the Cardiac Thin Filament

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Single amino acid mutations to the cardiac thin filament have been shown to cause genetic cardiomyopathies, a serious and sometimes deadly heart disorder. Experimental groups have extensively studied many of these mutations in an attempt to uncover the link between genotype and phenotype, but critical atomic level details cannot be determined by these methods. To address this gap, molecular dynamics (MD) simulations are utilized to model and study the changes in conformation and dynamics imposed by these single point mutations. The goal of this research is to move beyond identifying the effects mutations have on the cardiac thin filament, and to determine a set of guidelines to predict the clinical effects unstudied mutations will have on individuals. A set of twenty different clinically determined amino acid mutations located in cardiac troponin T and tropomyosin were chosen for analysis, and MD simulations were performed using the fully atomistic cardiac thin filament previously created by our group. From the simulations, conformation changes imposed by the mutation, both local and distant to the site of mutation, were computed. Steered molecular dynamics along with Jarzynski's equality were utilized to determine the changes in the free energy barrier of calcium binding and dissociation due to mutation. Coupling the changes in conformation and calcium dynamics data together, subsets of mutations were created to begin the process of determining guidelines for mutational predictions.

1302-Pos

Design of Biocompatible Liquid Cristal Elastomers Reproducing the Mechanical Properties of Human Cardiac Muscle

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Advanced materials, able to work as integrated actuators for the treatment of muscle injuries, could combine rapid and long-lasting intervention, which is the main goal in regenerative medicine. Among them, Liquid Crystalline Elastomers (LCEs) are biocompatible polymers able to reversibly deform in response to a given stimulus by generating movement. Once stimulated, LCEs can mimic muscle force production. However, so far their application in biology was limited by the slow response times and the reduced possibility to modulate tension levels during activation. Thanks to a screening of different monomeric formulations, a palette of biocompatible LCEs is prepared and precisely characterized in isometric conditions in terms of passive and active mechanical properties, showing improved muscle-like characteristic. Light responsive LCEs stimulated with increasing laser powers, develop progressively increasing active tensions with fast kinetics of activation and relaxation. Tension levels and contraction time course can be modulate to reproduce twitch contractions of cardiac samples from patients affected by specific diseases. As a proof of concept, a LCE-based device able to develop concentric pressure, as in cardiac chambers, was demonstrated opening for their use in the preparation of contraction assist devices.

1303-Pos

Human Beta-Cardiac Myosin Cardiomyopathy Mutations R712L and E497D Disrupt a Key Salt-Bridge in the Coupling Domain

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Mutations in the human beta-cardiac myosin gene (MYH7) are responsible for a large number of hypertrophic (HCM) and dilated (DCM) cardiomyopathies. Omecamtiv mecarbil (OM) is a small molecule drug that induces allosteric changes in beta-cardiac myosin activity and is in clinical trials for treatment of systolic heart failure. OM binds in a narrow cleft adjacent to the SH1 helix, relay helix, and converter domain of beta-cardiac myosin and interacts with residues involved in linking changes in the nucleotide pocket to rotation of the lever arm, known as the 'coupling region'. Cardiomyopathy mutations R712L and E497D disrupt a salt bridge between R712 and E497 that is centered in the OM binding pocket and plays a role in coupling the relay helix to a small beta-sheet of the converter domain. In the present study, we have characterized the effect of the mutations and OM on the unloaded shortening velocity of R712L and E497D beta-cardiac HMM mutants. The R712L mutation causes a 5-fold slowing of motility, and increases the IC50 of OM binding to $\sim$30 μM from 0.1 μM for the WT cHMM. OM also restores the velocity of R712L cHMM to 75% of WT cHMM velocity. On the other hand, the E497D mutation has little effect on the velocity, but increases the IC50 of OM to 4.5 μM. OM inhibits the velocity of E497D cHMM but only by 2.5-fold compared to over 25-fold for WT cHMM. In conclusion, the dramatic effect on motility (R712L) and impact on the IC50 of OM (R712L and E497D) suggest a central role of this buried salt bridge in coupling the nucleotide state to rotation of the converter domain.

1304-Pos

Isolating the Pathological Contribution of Detyrosinated Microtubules in Human Myocardial Mechanics

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Detyrosinated microtubules (dTyr MTs) buckle and bear load during myocyte contraction providing a viscoelastic resistance to myocyte shortening. The proliferation of dTyr MTs observed in heart failure increases myocyte viscoelasticity but the pathological consequences on myocyte and myocardial function remain to be quantitatively assessed. To determine whether suppressing dTyr MTs will reduce viscoelasticity and improve contractile dynamics proportional to the severity of disease, we utilize parallel genetic and pharmaceutical approaches to suppress MT dTyr in freshly isolated human cardiomyocytes and myocardial strips and measure active and passive mechanical properties. Compared to myocytes isolated from non-failing human hearts, suppression of dTyr MTs reduces viscoelasticity and increases unloaded shortening to a greater degree in failing cardiomyocytes. Manipulation of dTyr MT levels does not significantly impact excitation-contraction coupling and *de novo* mathematical modelling demonstrates that increased contractile dynamics can be attributed to the experimentally measured decrease in viscoelasticity. Preliminary results in myocardial strips from failing hearts display greater viscoelasticity than non-failing myocardium, with depolymerization of MTs in failing myocardium partially restoring passive mechanical properties. The results indicate that dTyr microtubules constrain myocyte contractile dynamics in heart failure. Moreover, the microtubules' impact in regulating myocardial mechanical properties at rates consistent with diastolic filling is evident in the presence of an intact extracellular matrix.

1305-Pos

Predicting and Preventing Myocardial Remodeling in a Murine Model of Dilated Cardiomyopathy

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In hearts with dilated cardiomyopathy (DCM), the ventricular walls become thin and weak, causing systolic dysfunction, and interventions to prevent the